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Fruiting and successions of ectomycorrhizal fungi

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Changes in the structure of fungal communities are often difficult to verify because hyphae, and other vegetative structures, can seldom be identified to species or even genus level without the preparation of pure cultures. The sequential appearance of reproductive structures in natural environments needs to be interpreted with care (Cooke & Rayner, 1984) because different fungi produce their reproductive structures at different ages (Harper & Webster, 1964). The presence of reproductive structures is always associated with the presence of mycelia, but their absence does not necessarily signify the absence of mycelia.

The (sheathing) ectomycorrhizas of birch (*Betula* spp.), pine (*Pinus* spp.) and spruce (*Picea* spp.), fortunately can be categorised by gross morphology and microscopical features such that the associated mycorrhizal fungi can be attributed at least to the genus (Last *et al.*, 1985). By coupling records of fruitbodies with direct observations of mycorrhizas it is possible, therefore, to study successional events with some degree of confidence for at least some tree species.

Concepts of fungal succession

Critical reviews of successions of saprotrophic fungi have recently been made by Frankland (1981) and Cooke & Rayner (1984). Earlier, Park (1968) drew analogies with the development of assemblages of higher plants and distinguished between (i) seral successions and (ii) substratum successions. The former equate to the series of changes leading to climax communities as has been described for fungi colonising sand dunes (Brown, 1958). Substratum successions, on the other hand, relate to events

on a smaller scale within a large environment; for example, on individual twigs or fallen leaves.

Seral successions doubtless occur among mycorrhizal fungi. For example, the mycorrhizal fungi associated with a seral succession of higher plants, progressing from herbaceous communities to climax woodland, will inevitably change from vesicular–arbuscular mycorrhizal fungi colonising herbs (Rose, 1980) to ectomycorrhizal fungi predominating on climax or sub-climax trees such as birch, pine, oak and beech. Successions can also occur in association with individual trees; these are substratum successions. In these instances, fungi differentially colonise roots at different times and at different distances from the tree base. Changes in populations of mycorrhizal fungi may be rapid, particularly in the early years of a tree's life. On a woodland scale, therefore, it is probable that both substratum and seral successions of mycorrhizal fungi occur, as trees exert strong influences both on their aerial and edaphic environments.

Successions of ectomycorrhizal fungi

Temporal aspects

Among mycorrhizal fungi, *Tricholoma fulvum* is characteristically associated with birch, *Suillus luteus* with pines, and *Suillus grevillei* with larch, but such associations are seldom exclusive, most tree species being associated with a spectrum of fungi, some host-specific and some of broad host-range as described in Chapter 2. A few tree species such as red alder (*Alnus rubra*), which develops in pure stands, tend to be more fungus-specific (Malajczuk, Molina & Trappe, 1982; Molina & Trappe, 1982). Apart from such exceptions, mycorrhizal successions, at least in previously treeless sites, appear to proceed through three broad phases: (1) a relatively narrow range of pioneer fungi of broad host-range develop in young stands, for example species of *Hebeloma, Laccaria* and *Inocybe*; (2) an increase in species-diversity occurs as the stand develops (see Table 12.1); (3) one or a few fungi become dominant in, or at least characterise, old stands, these fungi sometimes being host-specific and best adapted to the host in the particular edaphic conditions pertaining to a site. This view has evolved as a result of observations of mycorrhizal fungi associated with a variety of coniferous and broadleaved trees of different ages in many parts of the world (Chu-Chou, 1979; Chu-Chou & Grace, 1981; Mason *et al.*, 1982). A detailed study of the fungal fruitbodies arising annually in a young birch plantation (Table 12.1) revealed that certain species of *Inocybe, Hebeloma, Laccaria* and *Thelephora* appeared in the first four years after planting, followed largely in later years by species of *Lactarius, Leccinum, Cortinarius* and *Russula* (Mason *et al.*, 1982).

Table 12.1. *Succession of fruitbodies of proven or suspected ectomycorrhizal fungi appearing in a stand of birches planted at Bush estate, near Edinburgh (lat. 55°52'N) (after* Last et al., *1983)*

Year after planting	Fungi with fruit bodies occurring for the first time
1	Nil
	Hebeloma crustuliniforme
	Laccaria proxima
3	*Laccaria tortilis*
	Thelephora terrestris
4	*Hebeloma fragilipes*
	H. sacchariolens
	H. mesophaeum
	Inocybe lanuginella
	Lactarius pubescens
6	*Cortinarius* sp.
	Hebeloma leucosarx
	Hymenogaster tener
	Inocybe petiginosa
	Leccinum roseofractum
	L. scabrum
	L. versipelle
	Peziza badia
	Ramaria sp.
7	Other *Cortinarius* spp.
	Other *Hebeloma* spp.
	Lactarius glyciosmus
	Leccinum subleucophaeum
10	*Hebeloma vaccinum*
	Russula betularum
	R. grisea
	R. versicolor
14	*Laccaria laccata*
	Lactarius spinosulus
	Russula atropurpurea

Observations of fruitbodies beneath a number of other tree species growing in young stands revealed similarities to the populations of mycorrhizal fungi on young birch. J.H. Warcup (pers. comm.), studying four-year-old *Pinus contorta* in Scotland recorded *Hebeloma fragilipes, Thelephora terrestris, Amphimena byssoides, Rhizopogon luteolus* and *Suillus luteus* . This range of species is comparable to species of *Hebeloma, Rhizopogon* and *Suillus* found in young stands of *Pinus radiata, Pseudotsuga menziesii,* and *Eucalyptus* spp. growing in New Zealand (Chu–Chou, 1979; Chu-Chou & Grace, 1981, 1982, 1983) and *Picea sitchensis* in the UK (Thomas, Rogers & Jackson, 1983).

Watling (1981) similarly found species of *Laccaria* and *Hebeloma* pioneer fungi which colonised newly planted woodland and open land invaded by sapling trees; whereas in 19 established woodland sites in Perthshire he recorded fruitbodies of 45 species of *Cortinarius*, 38 of *Lactarius*, 57 of *Russula* and 7 of *Amanita* (Watling, 1984). Richardson (1970) found that the mycorrhizal flora of a mature (55-years-old) plantation of Scots pine (*Pinus sylvestris*) was dominated by *Russula* (*Russula emetica*) but also had significant numbers of fruitbodies of *Amanita* (*A. inaurata, A. rubescens* and *A. vaginata*) and *Lactarius* (*L. rufus* and *L. turpis*). In a stand of *Betula pendula* (72 years old), Miles (1985) found that 32% and 28% of the fruitbodies of mycorrhizal fungi were of *Amanita muscaria* and *Tricholoma columbetta*, respectively (Table 12.2).

Circumstantial evidence for the third phase mentioned above is provided by the relatively narrow range of fruitbodies often reported for ageing stands. Harvey, Larsen & Jurgensen (1976) found in a 250 year-old forest of Douglas fir and larch that roots of larch were associated with a single dominant mycorrhizal fungus, *Suillus cavipes*; whereas the fir roots were 'dominated' by *Russula brevipes*. Malajczuk *et al.* (1982) suggested that not only does a succession of mycorrhizal fungi occur in natural stands of eucalypts and *Pinus radiata*, but also that this succession, over time, tends towards dominance by host-specific fungi. Such uniformity does not occur in all mature woodlands, even if these contain only one tree species. The relict birchwood at Struan, Perthshire, is ancient and yet has a diverse community of ectomycorrhizal fungi (Fleming, Deacon & Last, 1986).

Spatial aspects

In addition to the age-related sequence, clear evidence of a spatial sequence of fruitbodies was noted in a young stand of birch (Ford, Mason & Pelham, 1980; Mason *et al.*, 1982). The mean distance of fruitbodies of different fungi from the bases of trees tended to increase in successive years, with later-appearing species usually fruiting, in the first instance, close to the trees, as did earlier appearing species before them (Ford *et al.*, 1980). However, while most species were associated with the fine roots in rings or arcs at particular distances from the trunk, some (e.g. *Laccaria tortilis*) developed a radial, spoke-like distribution as if following the main structural roots. The existence of these spatial distributions was corroborated by Deacon, Donaldson & Last (1983) who examined cores of soil at different distances from the bases of trees. For each tree, the predominant mycorrhizas typical of the early-fruiting species occurred furthest from the base.

Table 12.2. *Influence of age of natural stands of birch* (Betula pendula) *on the occurrence of fruitbodies of fungi known to form ectomycorrhizas with* B. pendula

	B. pendula age (yr)		
	20	29	72
	Occurrence of fruitbodies (per cent)		
Boletus impolitus	3	Nil	Nil
Cortinarius spp.	78	18	Nil
Hebeloma spp.	Nil	52	Nil
Lactarius pubescens	16	5	5
Russula atropurpurea	3	Nil	Nil
Amanita muscaria	Nil	8	32
Boletus edulis	Nil	3	Nil
Leccinum scabrum	Nil	3	5
Inocybe sp.	Nil	Nil	5
Laccaria amethystina	Nil	Nil	10
Lactarius tabidus	Nil	Nil	10
Russula aeruginea	Nil	3	5
Tricholoma columbetta	Nil	Nil	28
T. nudum	Nil	8	Nil

Data from Kerrow, near Cannich, Invernesshire (Miles, 1985).

The transition from nursery to outplanting sites

Several studies have involved seedlings transplanted from nursery or glasshouse environments to field sites, a transfer involving a major change of environment. Ectendomycorrhizal fungi, such as the 'E-strain' group, are typical of nursery soils but often disappear from the root system in the first few years after transplanting to field sites (Thomas *et al.*, 1983).

Teleomorphs of the 'E-strain' fungi are unknown but evidence suggests that these fungi may be ascomycetous, possibly species of *Geospora, Trichophaea, Sphaerosporella* or *Humaria* (Danielson, 1982). The Ascomycotina, as a group, has tended to be overlooked in studies of mycorrhizal successions because the fruitbodies are, for the most part, inconspicuous. Danielson (1984) and Danielson, Zak & Parkinson (1984) have drawn attention to the presence of ascomycetes associated with saplings in field soils. *Sphaerosporella brunnea*, for example, is a carbonicolous operculate discomycete that may play an important role in mycorrhizal succession in forest stands frequently subjected to fire (Danielson, 1984). It remains to be seen whether ascospores of this fungus have a degree of heat-resistance and are heat-activated, properties well recognised in *Rhizina undulata*, the important pathogen that causes 'group dying' of conifers on recently burned sites (Jalaluddin, 1967).

Effects of different genotypes

Different genotypes of one host may significantly modify at least the early stages of mycorrhizal successions, as shown for *Betula pubescens* (Last *et al.*, 1984). After planting rooted cuttings of two clones of *B. pubescens* (9.3D and 9.3G) in two soils, fruitbodies of *Hebeloma* spp. and *Inocybe petiginosa* developed around both clones in one soil; in the other, however, fruitbodies of *Laccaria tortilis* developed around 9.3G while *Hebeloma* spp. were found in association with 9.3D. It is not known if these early differences persist or affect the later stages of successions.

It seems that variation within the host is paralleled by similar variation within species of mycorrhizal fungi, as shown in *Amanita muscaria* by Mason (1975). Fries (1987) has recognised two morphologically distinct field populations of *Paxillus involutus* in Scandinavia; one associated with conifers and the other found with broadleaved trees such as birch.

Effects of different soils

Soil and site characteristics markedly influence the types of mycorrhizal fungi that develop on tree roots, though as yet there is insufficient information to enable generalisations to be made except where extreme differences occur. *Cenococcum geophilum* typically forms mycorrhizas in dry locations (Mikola, 1949; Pigott, 1982), whereas *Paxillus, Scleroderma* and *Pisolithus* are common on coal spoil and other mining wastes (Marx, 1980; Gardner & Malajczuk, 1985; Ingleby, Last & Mason, 1985). Gardner & Malajczuk (1985) noted that a fungal succession occurred in association with eucalypts on a bauxite mine, with *Pisolithus, Scleroderma* and *Laccaria* representing the early colonisers while *Cortinarius, Paxillus, Ramaria* and *Russula* spp. represented the later colonisers.

As trees develop on formerly treeless sites numerous alterations are caused to the soil. The environment becomes less favourable for litter breakdown and nutrient remobilisation by saprotrophic fungi (Swift, Heal & Anderson, 1979; Vogt *et al.*, 1983), and substantial amounts of organic matter accumulate. In mature forests, ectomycorrhizas tend to be concentrated in the upper soil horizons, particularly the litter and raw humus layers. In a Douglas fir/larch forest, Harvey *et al.* (1976) found 95% of active mycorrhizas associated with the organic soil fraction and only 5% with the underlying mineral soil.

Tyler (1985) noted the occurrence of fruitbodies in woodlands on a range of soil types and found that some mycorrhizal fungi were more prevalent in organic than in mineral soils. Whereas *Laccaria laccata*

fruited mainly at sites with small amounts of organic matter (<13%) *Amanita citrina, Cantharellus tubaeformis* and five *Cortinarius* spp. fruited mainly, if not exclusively, at sites with 30–93% organic matter. Such an observation is supported by Reddell & Malajczuk (1984) who grew jarrah (*Eucalyptus marginata*) seedlings in samples of mineral soil and forest litter taken from jarrah forests. Whereas black mycorrhizas (attributed to *Cenococcum geophilum*) were virtually confined to the mineral soils, white and brown mycorrhizas, attributed to basidiomycetes, were almost exclusively associated with seedlings growing in the litter.

Experiments have shown that mycorrhiza formation by *Paxillus involutus* is markedly affected by soil type. When spores of this fungus from fruitbodies occurring on coal spoil were added to samples of coal spoil and a brown earth in pots, birch seedlings developed many mycorrhizas of *Paxillus* in the coal spoil but only few mycorrhizas of this type (and on few seedlings) in the brown earth (Fox, 1986*a*). Last *et al.* (1985) found that *P. involutus*, after establishment on birch seedling roots in axenic culture, could persist and dominate the root system for at least a year after the seedlings were planted into pots of four contrasting (2 peat and 2 mineral) soils. However, by the end of the second year, *P. involutus* accounted for 44% of mycorrhizas in sphagnum peat of pH 3.4, and only 19% in a pH 5.4 mineral soil. In the same experiment, birch seedlings inoculated with *Hebeloma sacchariolens* had almost 100% of their roots tips colonised by this fungus after two years in both mineral soils (pH 5.1 and 5.4) and in sedge peat (pH 4.7) but the fungus completely disappeared from root systems in the first year in Sphagnum peat (pH 3.4). These observations from controlled experiments are corroborated by records made in the field. Thus, Dighton, Poskitt & Howard (1986) observed *Paxillus* and *Laccaria* in association with saplings of *Pinus contorta* and *Picea sitchensis* at a peat site, while Mason *et al.* (1982) found fruitbodies of *Hebeloma*, but not *Paxillus*, in conjunction with birch growing in a mineral soil.

In a detailed study made over four years, Fleming *et al.* (1984) grew *Betula pendula* for eight weeks from seed in two propagating soils; soils were taken from beneath the canopy ('mycorrhizal soil') of a birch tree and *c.* 11m from its periphery ('non-mycorrhizal'). Seedlings were then grown for a further two years in 'non-mycorrhizal' soil. Distinct assemblages of mycorrhizas developed on the seedlings during this time, with fruitbodies of *Hebeloma, Thelephora terrestris* and *Peziza badia* developing in association with the seedlings propagated initially in pots of 'non-mycorrhizal' soil, whereas *Inocybe lanuginella* and *Laccaria* fruited

in those propagated in 'mycorrhizal' soil. After two years of glasshouse cultivation, the seedlings were outplanted to a treeless site about 100m from where the propagating soils were collected. The periphery of the root zone of all plants became dominated by mycorrhizas of *Inocybe*. However, the two batches of plants still retained different types of mycorrhiza in the older parts of the root zone after a total of four years growth. The significance of this lies in the fact that the majority of mycorrhizal root tips occurred in these older regions of the root zone. Thus, the type of soil used for seedling propagation during the first few weeks of plant growth can have a significant influence on the mycorrhizal status of plants for at least four years.

Irrespective of host and soil effects, a broad distinction can be made between 'early' and 'late' fungal colonisers. The only important qualification that needs to be made is that these categories refer to mycorrhizal successions on previously tree-less sites, because from evidence presented in the next section it will be clear that early-stage fungi may be bypassed if seedlings develop in the root zone of old trees.

Experimental analysis of mycorrhizal succession

Succession in most, if not all, communities involves sequential colonisation and species-replacement.

Sequential colonisation

All fungi, whether early- or late-stage, that have so far been investigated, can establish mycorrhizas on very young seedlings from mycelial inocula in axenic culture (e.g. Mason, 1980); none has proved to be incompatible with seedlings. However, when equivalent inocula were added to unsterile (natural) soils, only early-stage fungi were able to establish mycorrhizas on seedling roots (Deacon *et al.*, 1983; Danielson, Visser & Parkinson, 1984; Last *et al.*, 1985). When Fox (1986*a*) added spores of late-stage fungi to unsterile soils, she found that birch seedlings either developed mycorrhizas of early-stage fungi, such as *Thelephora* or *Hebeloma*, from naturally occurring sources of inoculum, or the seedlings remained non-mycorrhizal. This suggests that competition from other mycorrhizal fungi is not the major factor preventing the establishment of late-stage fungi on seedlings in soil.

In glasshouse and growth chamber experiments, Fox (1983, 1986*a*) added large numbers of spores of different mycorrhizal fungi to pots of mineral soil from a treeless site and found that only the early-stage fungi in successions established mycorrhizas on birch seedlings in this soil. All

tested species of *Hebeloma (H. crustuliniforme, H. leucosarx* and *H. sacchariolens), Inocybe (I. geophylla, I. lacera, I. lanuginella)* and *Laccaria (L. proxima* and *L. tortilis)* were successful in establishing mycorrhizas from spores, whereas all species of *Cortinarius (C. bulbosus* and *C. debilitus), Lactarius (L. blennius, L. pubescens, L. rufus, L. spinosulus, L. turpis* and *L. vietus), Leccinum (Lecc. rosefractum* and *Lecc. scabrum)* and *Russula (R. cyanoxantha* and *R. grisea)* were unsuccessful. Fox's results suggest that under her conditions *genera* rather than individual species can be categorised as being able or unable to establish mycorrhizas from spores on seedling roots. Other workers have also reported major differences in the ability of fungi to establish mycorrhizas from spores in soil (Theodorou, 1971; Lamb & Richards, 1974*a,b*; Donald, 1975; Marx, 1976; Mullette, 1976; Marx, 1980). Marx (1980) reported that *Pisolithus* and *Thelephora* formed mycorrhizas from spores on pine seedlings in soil, whereas three species of *Amanita*, three of *Lactarius*, and *Suillus luteus, Russula emetica, Laccaria laccata* and *Paxillus involutus* did not.

A partial explanation of these results may lie in differences in spore germination between mycorrhizal fungi, though there have been few comparative studies of germination, and none that relates directly to mycorrhizal succession. On agar media, germination frequencies of 0.1% or less are reported for *Amanita muscaria, Leccinum scabrum, Lactarius helvus* and *Paxillus involutus*, even in the presence of activated charcoal or 'stimulatory' *Rhodotorula glutinis* (Fries, 1978). However, the relevance of this type of study is questionable since plant roots can stimulate the germination of basidiospores of mycorrhizal fungi (Melin, 1959; Birraux & Fries, 1981) and, also, because vegetative hyphae can stimulate germination, an effect that is highly taxon-specific for *Leccinum* spp. (Fries, 1981) but not for *Laccaria laccata* (Fries, 1983). It is not known whether homokaryotic mycelia derived from basidiospores can initiate mycorrhizal infection, or if dikaryotisation is necessary before infection can occur. Resolution of this question may be important because spores are the primary agents responsible for initiating mycorrhizal development – whether of early- or late-stage fungi – in sites that do not have a more or less continuous history of woodland cover. Nevertheless, it should be remembered that the behaviour of spore inocula in soils is paralleled by that of dikaryotic mycelial inocula produced in laboratory conditions and derived initially from fruitbody material.

In relation to mycorrhizal establishment, Fleming *et al.* (1986) found that 12 of 99 birch seedlings grown in pots of soil from an ancient

birchwood (Struan wood) developed mycorrhizas of *Amanita* and *Leccinum*, and many seedlings grown in soil samples from beneath fruitbodies of *Lactarius tabidus, Tricholoma fulvum* or *Cortinarius* spp. developed mycorrhizas of these respective fungi. These results contrast markedly with previous work on soil samples from a young birch stand (Deacon *et al.*, 1983) where only early-stage fungi developed mycorrhizas on birch seedlings, even if the soil samples were taken from beneath fruitbodies of late-stage fungi. They also contrast with results from a sweet chestnut coppice with oak standards, soil samples from which gave rise to only early-stage mycorrhizal types on birch seedlings in a glasshouse, whereas the predominant mycorrhizal type on tree roots in the field was typical of the Boletaceae (late-stage) (Fleming, 1983). The coppice woodland was at least 50 years old, as indicated by the size of the oak standards, but was on a shallow clay-with-flints soil overlying chalk and did not have a substantial accumulation of organic matter. The available but sparse evidence suggests that litter layers and humus-rich woodland soils may be more 'conducive' to the establishment of late-stage mycorrhizal types on seedlings than are predominantly mineral soils, a deduction possibly explaining the zonation of mycorrhizal types outwards from the tree base towards the periphery of a root system in spaced trees on previously tree-less sites.

Competition between primary mycorrhizal colonisers (early-stage), or at least niche exclusion by them, has been demonstrated in glasshouse studies with soils supplemented with spores (Fox, 1983, 1986*a*). When non-mycorrhizal birch, pine or spruce seedlings were grown in soil from a treeless site or in vermiculite-peat mixtures, mycorrhizas developed from spores of early-stage fungi. In contrast, when the soils were supplemented with spores of an early-stage inoculant fungus, such as *Hebeloma, Laccaria* or *Inocybe*, the same number of mycorrhizas developed as in the non-supplemented soil but were usually solely attributable to the inoculant fungus, which thus excluded the 'contaminants'. The same was observed when axenically grown birch seedlings, colonised by mycelial cultures of *Hebeloma sacchariolens*, were transferred to unsterile soils (Mason *et al.*, 1983). This ineffectiveness of introduced inoculum may have implications for the commercial inoculation of trees with mycorrhizal fungi.

Species replacement

The possibility that early-stage mycorrhizal fungi facilitate the subsequent establishment of late-stage fungi was investigated by Fleming (1985). Birch seedlings were inoculated with *Hebeloma sacchariolens* or

Laccaria proxima, or left uninoculated, before planting in a brown earth supplemented with mycelial inocula of *Amanita muscaria*, *Leccinum scabrum* or *Lactarius pubescens*. For the 441 d duration of the experiment, no mycorrhizas of *Amanita* or *Leccinum* developed. *Lactarius* mycorrhizas developed on roots, but independently of inoculation with *L. pubescens*, suggesting that they arose from naturally-occurring inoculum. At the end of the experiment *Lactarius* mycorrhizas represented up to 30% of the total number of mycorrhizas occurring on the originally non-mycorrhizal seedlings and those inoculated with *Hebeloma*, whereas none occurred on seedlings initially mycorrhizal with *Laccaria*. There was, therefore, no evidence of facilitated replacement of early-stage fungi by late-stage fungi, but there was evidence of inhibition of the development of the late-stage fungus, *Lactarius pubescens*, by *Laccaria* previously established on the root systems. In some respects these results parallel those of Fleming *et al.* (1984) who found that *L. pubescens* from natural inoculum sources colonised birch saplings if these had previously become colonised by *Hebeloma* sp. but not if they were colonised by *Inocybe* sp.

Two other experimental approaches have yielded information relating to replacement phenomena. In the first (Fleming, 1985), mycelial cultures of three early-stage fungi (*Paxillus involutus*, *Hebeloma sacchariolens* and *Thelephora terrestris*) were either inoculated onto seedlings in aseptic conditions (primary inoculants) or added to soil as 'secondary inoculants'. The seedlings were then planted in soil so that the different combinations of primary and secondary inoculants were compared together with uninoculated controls. In such an experiment in which two 'soils' were used – a brown earth and a peat – *Paxillus* did not persist as a primary inoculant in any soil or soil treatment, even if no secondary inoculant was present. Instead, in the absence of a secondary inoculant the root systems were colonised by *Thelephora* in brown earth and *Inocybe lacera* in peat, from presumably low levels of naturally-occurring inoculum. *Paxillus* colonised all seedlings in all treatments if it was used as a secondary inoculant, and it then replaced the primary inoculants. In contrast to *Paxillus*, *Thelephora* persisted as a primary inoculant in both soils in the absence of secondary inoculants, but was replaced by *Paxillus* secondary inoculum in both soils and by *Hebeloma* secondary inoculum in brown earth. Yet another pattern was displayed by *Hebeloma*, which persisted as a primary inoculant in brown earth and not in peat, but was replaced by *Paxillus* (secondary inoculant) in both soils. Thus, in addition to demonstrating the over-riding importance of soil type, the experiment

showed unexpectedly that *Paxillus involutus* was a successful colonist when added to soil as a secondary inoculant but failed to persist when inoculated to seedlings as a primary inoculant. *Hebeloma* and *Thelephora* experienced no such difficulty as primary inoculants in soils favourable to their development.

In the second approach, non-mycorrhizal birch seedlings were planted among the roots of old trees (Fleming, 1983, 1984; Fleming *et al.*, 1986). The early stages of mycorrhizal succession were bypassed, because even young seedlings developed late-stage mycorrhizas or a mixture of early- and late-stage types. However, if the seedlings were planted into soil which had previously been cored or trenched to separate the roots in it from contact with the parent tree, then late-stage mycorrhizal fungi developed poorly, if at all, on the seedling roots. It seems, therefore, that the ability of late-stage fungi to colonise the roots of seedlings growing in unsterile soils is enhanced if they have uninterrupted links with their food bases represented by their 'parent' tree, a situation comparable to the behaviour of rhizomorphs of the pathogen *Armillaria mellea* and strands of *Serpula lacrymans*, the dry rot fungus (Garrett, 1956).

In terms of replacement, fungi that form mycelial strands or copious networks of unaggregated hyphae from the mycorrhizal sheath might be expected progressively to dominate root systems once they are established in a succession. Many late-stage mycorrhizal fungi do, indeed, produce mycelial strands, which may serve roles in addition to infection, such as the transport of water and mineral nutrients (Read, Francis & Finlay, 1985). In contrast, many early-stage fungi do not produce conspicuous mycelial strands and, instead, rely on spore inocula for colonisation of new flushes of roots. Persistence of some of these early-stage fungi may be facilitated by the production of sclerotia, as in *Hebeloma sacchariolens* (Fox, 1986*b*) and *Paxillus involutus* (Fox, 1986*c*).

Concluding remarks

For long periods the existence and value of mycorrhizas was taken for granted by foresters. From observations of fungal fruitbodies, and more recently supported by inoculation experiments, it has been possible to identify some of the factors which together control a well-ordered, age-related sequence (succession) of mycorrhizal fungi colonising the roots of trees. With the recognition of the phenomena of succession and host specificity (Chapter 2), and the need to take account of the influence of soil type, it is clear that the stage is set for a rational approach to the manipulation of mycorrhizal successions in forests. Bearing in mind the

increasing areas felled and replanted, it is now imperative to broaden our focus to include the ecology of mycorrhizal fungi in second-rotation forests, which may not be the same as in the first rotation.

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